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Note

Determination of carboxylic acids in the fruits of *Laurocerasus officinalis* Roem. and its cultivars

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Twenty-two compounds have been identified from ether extracts of the fruits of cultivars (Oxygemmis, Globigemmis and Angustifolia) and the wild form of *Laurocerasus officinalis* Roem. These are lactic acid, 2-furancarboxylic acid, benzyl alcohol, benzoic acid, α-hydroxy-β-methylvaleric acid, benzeneacetic acid, succinic acid, methylsuccinic acid, 2-butenedioic acid, α-hydroxybenzeneacetic acid, 5-hydroxymaltol, hydroxysuccinic acid, cinnamic acid, α-hydroxybenzene propanoic acid, 4-hydroxybenzoic acid, 4-hydroxybenzene acetic acid, vanillic acid, azelaic acid, 3,4-dihydroxybenzoic acid, *p*-coumaric acid, hydroxysuccinic acid benzoate and caffeic acid. Hydroxysuccinic acid, hydroxysuccinic acid benzoate and benzoic acid are the major compounds in these fruits. Benzyl alcohol, 4-hydroxybenzene acetic acid and *p*-coumaric acid are found only in the fruits of the wild form of *Laurocerasus officinalis* Roem. 5-Hydroxymaltol has been identified only in the fruits of Angustifolia.

Laurocerasus officinalis Roem. (syn: *Prunus laurocerasus* L.) (cherry laurel) is an evergreen plant usually up to 6 m in height[1]. Distribution of the wild form and its related cultivars and the botanical features of fruits of *L. officinalis* have been described in detail in our previous papers[2,3]. In recent years the fresh and dried fruits of the cultivated plants, being used for making jam, dried fruit pulp, marmalade and adorning alcoholic drinks, are the newly introduced potential food sources in Turkey[3,4]. This is the first report about the carboxylic acid compositions in the wild and cultivated plants of cherry laurels found in Turkey. The cultivars Oxygemmis and Globigemmis are new in Turkey[5]. The compositions of carboxylic acids in the fruits of cultivars and the wild form of *L. officinalis* were compared. A more detailed knowledge of variability in the carboxylic acid contents of the cultivars could be the desirable feature for selecting *L. officinalis* with improved nutritional quality.

The retention times and percentage area of peaks in TIC are given in **Table I**. Total ion current (TIC) chromatograms of each extract of four different species of cherry laurels are presented in **Figures 1-4**. The peak numbers of compounds on the chromatograms that belong to each fruit extract are given in the table. The percentage compositions of carboxylic acids were estimated on the basis of integrated TIC chromatograms generated during each GC-MS run. We have used the percentage area of the peaks in total ion current for relative quantification of compounds in the analyzed fruit extracts.

In the ether extracts of the fruits of cultivars and wild form of cherry laurels, 22 different compounds were identified by GC-MS analysis. In these fruit extracts, 17 compounds for Oxygemmis, 14 compounds for Globigemmis, 18 compounds for Angustifolia and 16 compounds for the wild form were identified. The compound levels which were less than or equal to 0.1% are not given in the table. Hydroxysuccinic, hydroxysuccinic acid benzoate and benzoic acids were the major compounds in these fruits. Benzyl alcohol, 4-hydroxybenzeneacetic acid and *p*-coumaric acid were identified only in the fruits of wild form while 5-hydroxymaltol was found in the fruits of *Angustifolia.* Hence, further detailed and careful studies on the carboxylic acid contents and other metabolites will help in the assessment of adequate chemotaxonomic information and provide helpful

Table I—Carboxylic acids identified in the fruits of cultivars and the wild form of *L. officinalis* Roem

Peak no.	Retention time	Compound	% area of peak in TIC			
			L. officinalis cvs.			*L. officinalis*
			Oxygemmis	*Globigemmis*	*Angustifolia*	wild form
1	4.218	Lactic acid	0.33	1.65	0.38	0.58
2	4.961	2-Furancarboxylic acid	–*	0.27	–	0.58
3	5.407	Benzyl alcohol	–	–	–	4.11
4	6.825	Benzoic acid	8.86	18.2	18.24	26.69
5	7.147	α-Hydroxy-β-methylvaleric acid	0.37	0.89	0.65	–
6	7.564	Benzeneacetic acid	3.14	3.35	1.61	1.46
7	8.122	Succinic acid	5.45	4.77	4.08	2.18
8	8.360	Methyl succinic acid	0.58	–	0.34	–
9	8.779	2-Butenedioic acid	4.91	6.72	8.45	1.38
10	10.731	α-Hydroxybenzeneacetic acid	7.09	10.21	8.21	2.59
11	10.816	5-Hydroxymaltol	–	–	0.40	–
12	11.271	Hydroxysuccinic acid	23.80	25.27	20.24	11.62
13	11.399	Cinnamic acid	0.37	0.55	0.88	–
14	12.359	α-Hydroxybenzene propanoic acid	0.54	0.50	0.46	0.55
15	12.939	4-Hydroxybenzoic acid	1.39	0.70	0.52	0.65
16	13.057	4-Hydroxybenzene acetic acid	–	–	–	0.32
17	14.831	Vanillic acid	4.46	1.70	2.77	3.27
18	15.338	Azelaic acid	0.38	–	0.15	–
19	15.789	3,4-Dihydroxybenzoic acid	0.86	–	0.23	1.13
20	17.078	*p*-Coumaric acid	–	–	–	0.50
21	17.963	Hydroxysuccinic acid benzoate	25.41	16.69	19.14	30.41
22	19.709	Caffeic acid	0.37	–	0.29	0.33

*Not determined

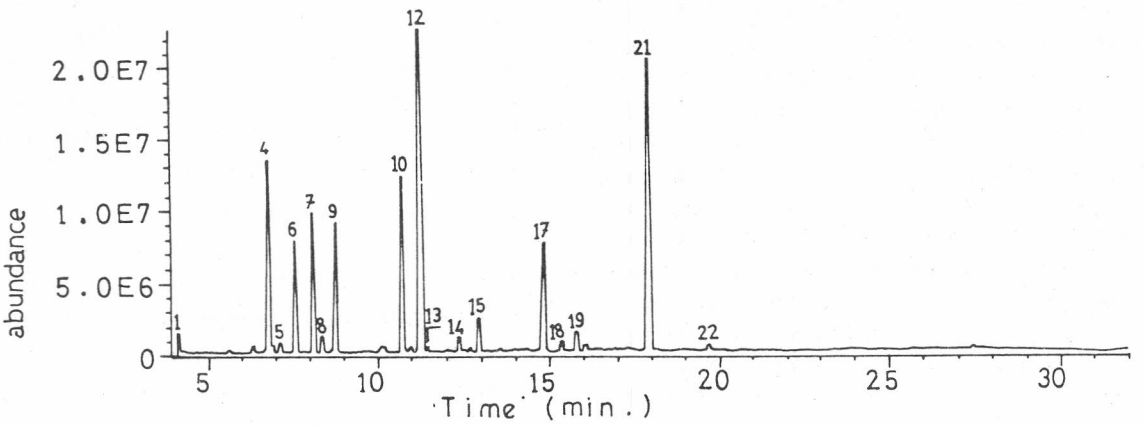

Figure 1—Total ion chromatogram of carboxylic acids from the fruits of cv. Oxygemmis of *L. officinalis* Roem. The peak numbers are given in Table I.

suggestion to correlate the chemical composition with geographical, climatic and botanical parameters.

Experimental Section

Extraction of carboxylic acids from cherry laurel fruits. The well-ripened fruits of cherry laurel cultivars (cv. Oxygemmis, Globigemmis and Angustifolia) and the wild form were collected randomly from their natural habitats in early morning in mid-August 1995 from young trees in the vicinity of Trabzon (Turkey). The fruit mesocarps were dried at 60°C under *vacuo* for 24 hr.

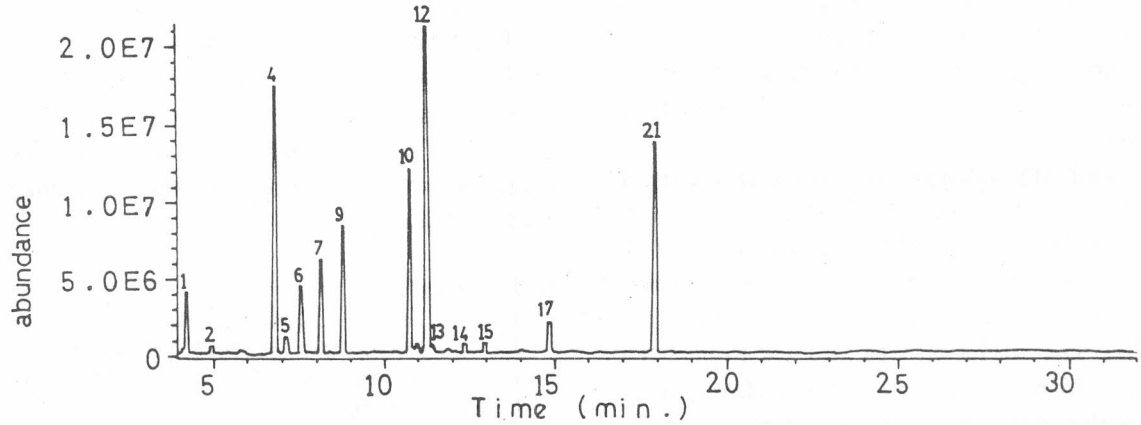

Figure 2—Total ion chromatogram of carboxylic acids from the fruits of cv. *Globigemmis* of *L. officinalis* Roem. The peak numbers are given in Table I.

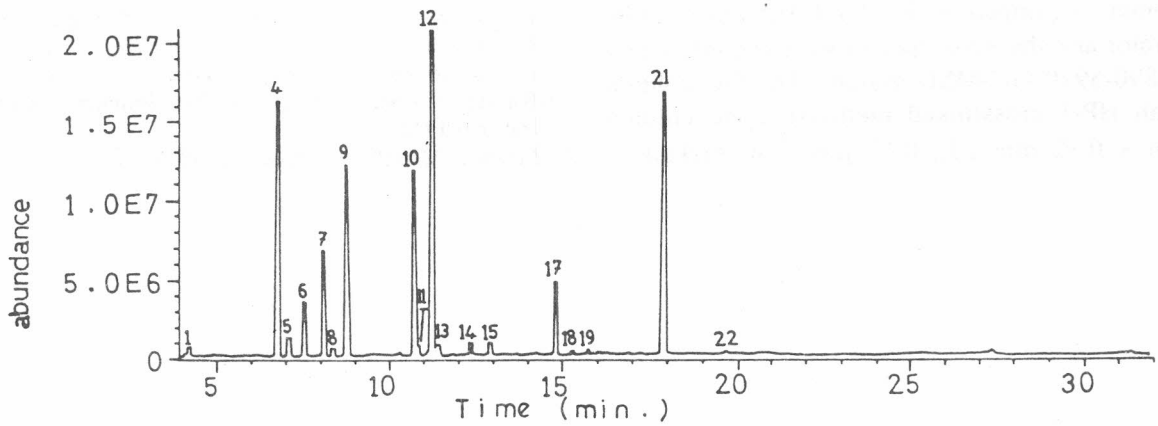

Figure 3—Total ion chromatogram of carboxylic acids from the fruits of cv. Angustifolia of *L. officinalis* Roem. The peak numbers are given in Table I.

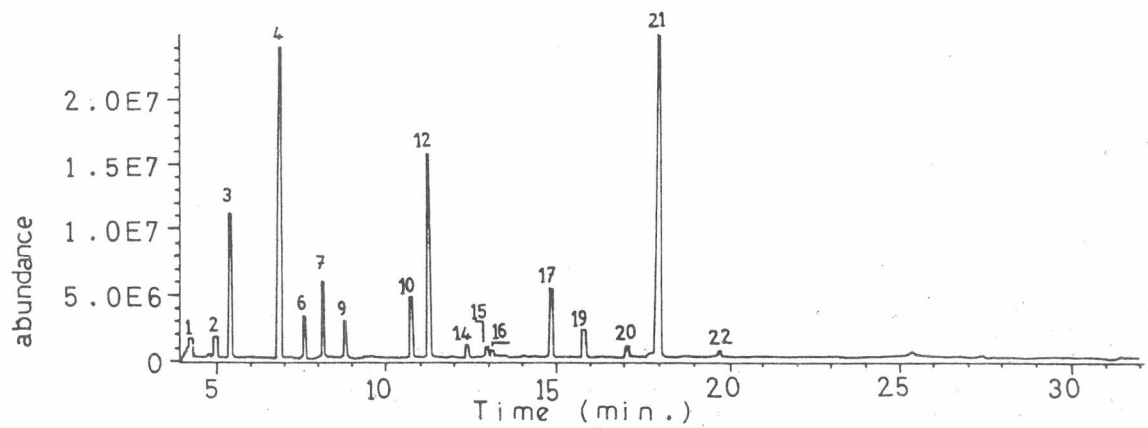

Figure 4—Total ion chromatogram of carboxylic acids from the fruits of the wild form of *L. officinalis* Roem. The peak numbers are given in Table I.

The extraction of carboxylic acids was made according to the method of Zapata and Mcmillan[6]. For the extraction, 20 g ground fruit sample (mesocarps) was used in triplicate and the extracts of triplicates were subjected to GC-MS analysis separately.

GC and MS analysis of carboxylic acids. The extracts were dissolved in pyridine and converted to their trimethylsilyl (TMS) derivates by adding a 1:1 (v/v) solution of bis-(trimethylsilyl)trifluoro-acetamide (BSTFA) and trimethylchlorosilane (TMCS). The silylation was allowed to proceed at 70°C for 15 min. Thereafter the silylated samples were analysed by gas chromatography using flame ionisation detector (GC-FID), and by combined gas chromatography-mass spectrometry (GC-MS). The GC analysis was carried out on a Varian 3300 instrument equipped with Merck-Hitachi D-2000 integrator and the mass spectra were recorded on a HP 5890-5970 GC-MSD system. The GC-column was an HP-1 crosslinked methylsiloxane column (25 m × 0.32 mm i.d., 0.17 μm film thickness). Hydrogen (helium in GC-MS) was used as the carrier gas at a flow rate of 55 cm/s. The column oven temperature was programmed starting from 60°C to 290°C at 8°C/min heating rate and the injector and FID temperatures were held at 260°C and 300°C, respectively. Identification of compounds was based on published reference mass spectral library.

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